

REEP1 siRNA (h): sc-94388

BACKGROUND

Transport of G protein-coupled receptors (GPCRs) to the cell surface membrane is critical for receptor-ligand recognition. Mammalian GPCR odorant receptors (ORs), when heterologously expressed in cells, are poorly expressed on the cell surface. REEP1 (receptor expression-enhancing protein 1), is a 201 amino acid multi-pass mitochondrion membrane protein that belongs to the DP1 family. REEP1 interacts with odorant receptor proteins and may enhance the cell surface expression of odorant receptors. Mutations in the REEP1 gene are the third most common cause of hereditary spastic paraplegia (HSP) after spastin and atlastin gene mutations. Mutations in the REEP1 gene also cause spastic paraplegia autosomal dominant type 31, a neurodegenerative disorder. The REEP1 gene is conserved in chimpanzee, canine, bovine, mouse, rat, chicken, zebrafish, *A. thaliana* and rice, and maps to human chromosome 2p11.2.

REFERENCES

1. Saito, H., et al. 2004. RTP family members induce functional expression of mammalian odorant receptors. *Cell* 119: 679-691.
2. Online Mendelian Inheritance in Man, OMIM™. 2005. Johns Hopkins University, Baltimore, MD. MIM Number: 609139. World Wide Web URL: <http://www.ncbi.nlm.nih.gov/omim/>
3. Züchner, S., et al. 2006. Mutations in the novel mitochondrial protein REEP1 cause hereditary spastic paraplegia type 31. *Am. J. Hum. Genet.* 79: 365-369.
4. Behrens, M., et al. 2006. Members of RTP and REEP gene families influence functional bitter taste receptor expression. *J. Biol. Chem.* 281: 20650-20659.
5. Beetz, C., et al. 2008. REEP1 mutation spectrum and genotype/phenotype correlation in hereditary spastic paraplegia type 31. *Brain* 131: 1078-1086.
6. Tzschach, A., et al. 2009. Interstitial deletion 2p11.2-p12: report of a patient with mental retardation and review of the literature. *Am. J. Med. Genet. A* 149A: 242-245.
7. Hewamadduma, C., et al. 2009. New pedigrees and novel mutation expand the phenotype of REEP1-associated hereditary spastic paraplegia (HSP). *Neurogenetics* 10: 105-110.

CHROMOSOMAL LOCATION

Genetic locus: REEP1 (human) mapping to 2p11.2.

PRODUCT

REEP1 siRNA (h) is a pool of 3 target-specific 19-25 nt siRNAs designed to knock down gene expression. Each vial contains 3.3 nmol of lyophilized siRNA, sufficient for a 10 μ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see REEP1 shRNA Plasmid (h): sc-94388-SH and REEP1 shRNA (h) Lentiviral Particles: sc-94388-V as alternate gene silencing products.

For independent verification of REEP1 (h) gene silencing results, we also provide the individual siRNA duplex components. Each is available as 3.3 nmol of lyophilized siRNA. These include: sc-94388A, sc-94388B and sc-94388C.

STORAGE AND RESUSPENSION

Store lyophilized siRNA duplex at -20° C with desiccant. Stable for at least one year from the date of shipment. Once resuspended, store at -20° C, avoid contact with RNases and repeated freeze thaw cycles.

Resuspend lyophilized siRNA duplex in 330 μ l of the RNase-free water provided. Resuspension of the siRNA duplex in 330 μ l of RNase-free water makes a 10 μ M solution in a 10 μ M Tris-HCl, pH 8.0, 20 mM NaCl, 1 mM EDTA buffered solution.

APPLICATIONS

REEP1 siRNA (h) is recommended for the inhibition of REEP1 expression in human cells.

SUPPORT REAGENTS

For optimal siRNA transfection efficiency, Santa Cruz Biotechnology's siRNA Transfection Reagent: sc-29528 (0.3 ml), siRNA Transfection Medium: sc-36868 (20 ml) and siRNA Dilution Buffer: sc-29527 (1.5 ml) are recommended. Control siRNAs or Fluorescein Conjugated Control siRNAs are available as 10 μ M in 66 μ l. Each contain a scrambled sequence that will not lead to the specific degradation of any known cellular mRNA. Fluorescein Conjugated Control siRNAs include: sc-36869, sc-44239, sc-44240 and sc-44241. Control siRNAs include: sc-37007, sc-44230, sc-44231, sc-44232, sc-44233, sc-44234, sc-44235, sc-44236, sc-44237 and sc-44238.

GENE EXPRESSION MONITORING

REEP1 (A-8): sc-393242 is recommended as a control antibody for monitoring of REEP1 gene expression knockdown by Western Blotting (starting dilution 1:200, dilution range 1:100-1:1000) or immunofluorescence (starting dilution 1:50, dilution range 1:50-1:500).

To ensure optimal results, the following support reagents are recommended: 1) Western Blotting: use m-IgG κ BP-HRP: sc-516102 or m-IgG κ BP-HRP (Cruz Marker): sc-516102-CM (dilution range: 1:1000-1:10000), Cruz Marker™ Molecular Weight Standards: sc-2035, UltraCruz® Blocking Reagent: sc-516214 and Western Blotting Luminol Reagent: sc-2048. 2) Immunofluorescence: use m-IgG κ BP-FITC: sc-516140 or m-IgG κ BP-PE: sc-516141 (dilution range: 1:50-1:200) with UltraCruz® Mounting Medium: sc-24941 or UltraCruz® Hard-set Mounting Medium: sc-359850.

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor REEP1 gene expression knockdown using RT-PCR Primer: REEP1 (h)-PR: sc-94388-PR (20 μ l). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

RESEARCH USE

For research use only, not for use in diagnostic procedures.

PROTOCOLS

See our web site at www.scbt.com for detailed protocols and support products.